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Technical Report

CRDLR 3023

Lethality of VX in Rats at High and Low Temperature

by

H. M. Frankel
J. S. Wiles

August 1960



ARMY CHEMICAL CENTER, MD

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CHEMICAL RESEARCH AND DEVELOPMENT LABORATORIES
Army Chemical Center, Maryland

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FOREWORD

This work was conducted under Task 4C08-02-023-01, Basic and Applied Physiology (U), and Task 4C08-02-023-04, Toxicology of CW Agents (U). The work was started in January 1959 and completed in July 1959. The experimental data are recorded in notebook MN-1127.

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DIGEST

The object of this research was to compare the lethality of VX injected intravenously and applied percutaneously at low, normal, and high environmental temperatures.

The conclusions reached are as follows:

1. Acute exposure of rats at 2° or 34°C increased the toxicity of VX administered intravenously or percutaneously when compared with exposure at 26°C.
2. The effect of temperature on lethality of VX is more marked with percutaneous application than with intravenous injection.

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LETHALITY OF VX IN RATS AT HIGH AND LOW TEMPERATURE

I. INTRODUCTION.

Acute exposure to high or low environmental temperature modifies the toxicity of anticholinesterase compounds in animals. Oberst et al.¹ have shown that at a temperature ranging from 32° to 38°C, the $\overline{\text{LCt50}}$ for GB on the skin of monkeys was 59% less than when the animals were exposed at a temperature ranging from 21° to 27°C. These findings were supported by the work of Frankel and Craig.² McPhail³ has reported that the LCt50 of GB inhaled by rats exposed at from -18°C to 0°C was from 12% to 86% less than the value obtained at from 13° to 16°C. However, McPhail and Buckholz⁴ showed that the LD50 of GB on the skin of pigs was 50% greater during exposure at 3°C than at 16°C. Similarly, in monkeys the LD50 on the skin was 50% greater at 3°C than at from 23° to 28°C.

Marzulli and Callahan⁵ were of the opinion, from blood cholinesterase changes in goats, that GB was more rapidly absorbed during the spring and summer seasons than in the winter. It has been reported⁶ that the Lt50 for VX on the skin of cats and rabbits was shorter at 32°C and longer at 2°C than at 26°C. These effects were modified by acclimation and the site of application.⁷

That penetration of the skin is not the only factor in these changes in percutaneous toxicity is seen from the following. Exposure to high or low environmental temperature will decrease the intravenous LD50 of GB in rats.⁸ Streicher⁹ demonstrated a decreased LD50 for DFP injected intraperitoneally in mice exposed at 4°C. Also, 30 μl of GB/kg injected intramuscularly killed more monkeys exposed at 38°C than at 25°C.² Baetjer and Smith¹⁰ showed, in mice injected intraperitoneally and intravenously, that exposure at either 16° or 36°C increased the toxicity of parathion over the toxicity at room temperature.

The reason for the existence of different tolerances at various environmental temperatures is not known. It has been suggested that the rate and efficiency of absorption may be factors. Ainsworth¹¹ applied GB containing radioactive phosphorus to rabbit skin and demonstrated that the concentration of labeled phosphorus in the body was inversely related to the skin temperature. The present study was designed to isolate the contribution of penetration through skin from the other factors that produce changes in toxicity with temperature.

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II. INVESTIGATIONAL PROCEDURES.

Male albino rats, weighing from 167 to 380 gm, were exposed to controlled temperatures of 2°, 26°, or 34°C for 20 hours before and for 24 hours after application of VX. They were housed in community cages 24 by 18 by 18 in. containing 10 to 12 animals. The agent was injected either directly into the femoral vein or applied as a drop to the surface of the skin on a clipped area in the middle of the back. The animals were selected at random for the dosage and temperature treatment they were to receive. Rats that were given agent on the back were placed in individual compartments 4 by 8 by 4 in. to decrease the opportunity of their licking the agent. Dilutions of the agent were made immediately before use. The first two dilutions were in propylene glycol and the final dilution for intravenous injection was in isotonic saline. The final dilution for percutaneous application was in isopropanol. Dosage was expressed in microliters of undiluted agent, sp gr 1.004.

All animals were injected at room temperature and were returned to the controlled temperature within a few minutes after injection. Application of the agent percutaneously was carried out at the controlled temperature. Agent was given to animals at all three temperatures during the same test period. Deaths were recorded during the following 24 hours. Regression slope and LD50 were calculated by the Bliss method.¹² A "p" value of 0.05 was used to test significance.

III. RESULTS.

Exposure to 2° or 34°C tended to increase the toxicity of a given dose compared with exposure at 26°C when the VX was administered intravenously or percutaneously (see figure). The intravenous LD50's at 2° and 34°C were 86% and 59%, respectively, of the doses required at 26°C. The percutaneous doses were 30% and 35% for 2° and 34°C, respectively, of the dose required at 26°C, as shown in table. The percutaneous traversal of VX, that is, the ratio of $\frac{\text{LD50, intravenous}}{\text{LD50, percutaneous}}$ has been used by Marzulli¹³ for a measure of agent effectiveness. In the present study this ratio was 0.196 at 2°C, 0.070 at 26°C, and 0.116 at 34°C.

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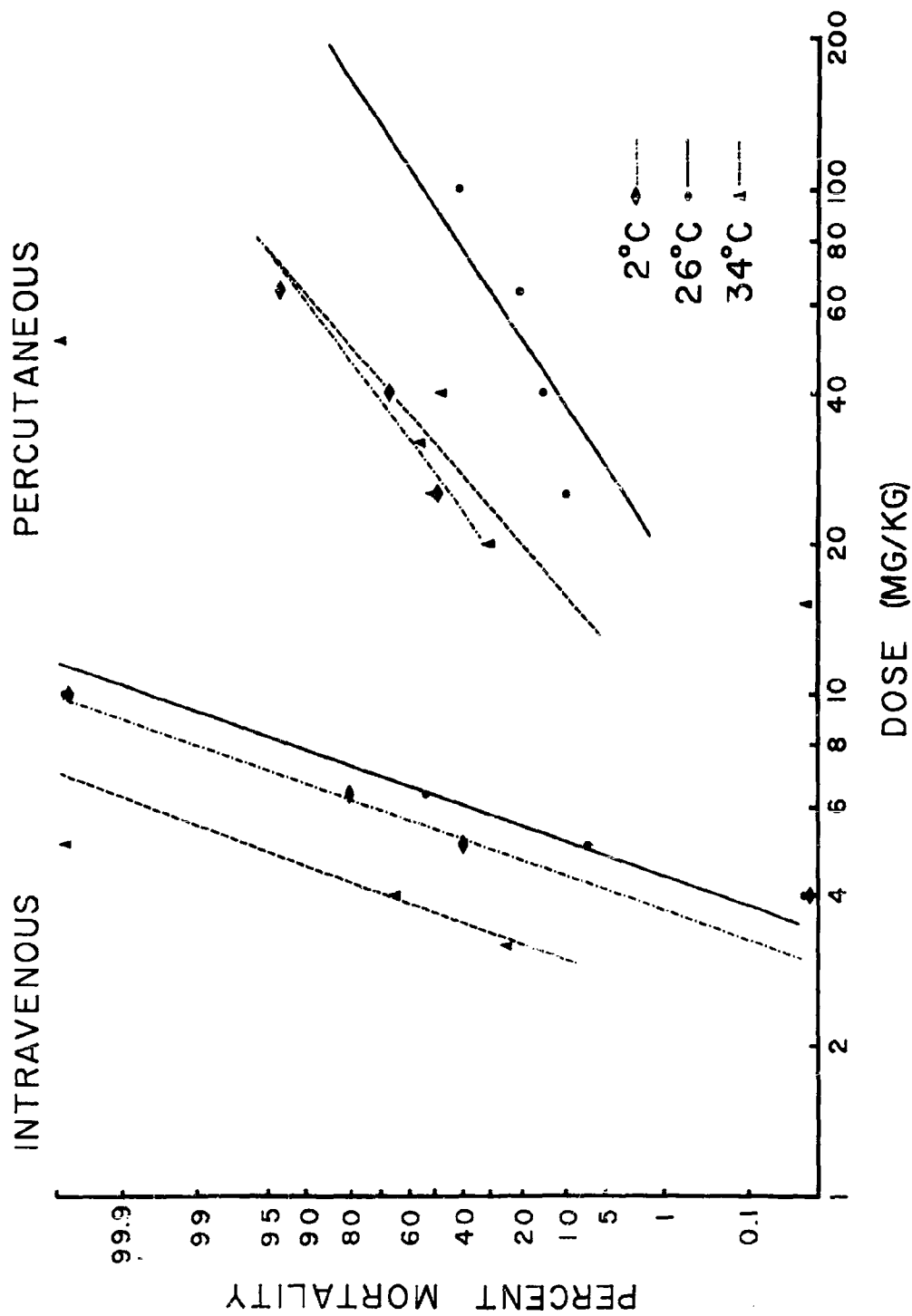


TABLE
LETHALITY OF VX IN RATS AT VARIOUS TEMPERATURES

Route of administration	Environmental temperature	LD50	95% Confidence limits	Number of rats	Regression slope
	°C	μl/kg			
Intravenous	2	5.4	4.9-5.9	35	14.55
Intravenous	26	6.3	5.8-7.4	45	13.86
Intravenous	34	3.7	3.3-4.1	22	15.23
Percutaneous	2	27.6	20.0-38.1	45	3.71
Percutaneous	26	91.7	70.4-119.6	65	3.34
Percutaneous	34	31.9	26.5-38.2	89	4.42

IV. DISCUSSION.

At 26°C, 91.7 μl/kg of VX must be placed on the skin of rats to produce the same effect as 6.3 μl/kg injected into the blood stream. The difference, or 85.4 μl/kg, represents agent that was lost or inactivated somewhere along the line from the surface of the skin to the blood stream. Either this difference or the ratio between the percutaneous and the intravenous doses may provide a measure of the resistance or penetrability of the skin or of efficiency of absorption. From the percutaneous traversal ratios described in the results it appears that agent is more effectively absorbed through the skin of rats exposed at 2° and 34°C than at 26°C.

The mechanism responsible for the change in efficiency is not clear. It cannot be circulation alone, because in the cold circulation in the skin tends to decrease. It may be a combination of factors, such as, the time the agent is in contact with the skin and circulation to the area. Thus, in the cold, time on the surface may increase and circulation decline; in the heat, time may decrease and circulation to the area increase. In this report it has been the authors' observation that time to death was longer in the cold than at the highest temperature. A measurement of circulation in the area at the time the agent is in contact with the skin would help in the interpretation of these results.

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V. CONCLUSIONS.

The conclusions reached are as follows:

1. Acute exposure of rats at 2° or 34°C increased the toxicity of VX administered intravenously or percutaneously when compared with exposure at 26°C.

2. The effect of temperature on lethality of VX is more marked with percutaneous application than with intravenous injection.

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ECBC Documents for Downgrading/Change in Distribution

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